

# Identification of Identical Twins and Mutation rate in pigs

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## ASL-R662

### Summary and Implications

A Berkshire X Yorkshire three-generation population from a quantitative trait loci (QTL) study was used to determine whether identical twin births occurred in pigs. The population consisted of 65 families with 525 F2 individuals. Genotype data at 125 microsatellite marker loci were examined for all F2 animals. One pair of female twins was found in the population. This is a unique result because there have been no published reports to date of twin births in pigs. In addition, three mutations of the microsatellite markers used in the QTL study were discovered out of the 134,565 meioses that occurred. This gives an overall mutation rate for the population of  $2.23 \times 10^{-5}$ . This mutation rate is similar to previously published data.

### Introduction

Quantitative trait loci (QTL) studies are a way to scan the genome of a species for regions that may affect economically important traits. Animals are genotyped using genetic markers. The most commonly used markers are microsatellites. Microsatellites are sections of DNA made up of repeating short nucleotide sequences. Genotyping animals by using microsatellites is an expensive process. So, maximizing the amount of information obtained from a set of genotype data is advantageous.

To date there have been no confirmed cases of identical twin pig births. Because pigs are a polytocous species, identical twins can only be detected by analyzing animals at the genetic level. Twin pig embryos that were confirmed by DNA fingerprinting have been reported (1). However, competition between embryos of different sizes or different stages of development is widely known to occur (2,3). Estimates of twinning rate taken during gestation may not accurately reflect the birth rate of twins.

Microsatellites are able to detect single base pair changes in DNA, which makes them ideal for comparing the genomes of individuals. Also, studying the rate at which these microsatellites mutate helps further the understanding of the evolution, generation, and persistence of simple repeated sequences in DNA (4).

The objectives of this study were to determine the existence and/or frequency of identical twin births in pigs by using marker data from a QTL linkage analysis study and to calculate a mutation rate in the resource population for those porcine microsatellite markers.

### Materials and Methods

The population for the QTL study was created by mating two Berkshire males with nine Yorkshire females and then crossing their offspring to produce 65 different families. All individuals,  $n=572$ , were genotyped by GeneSeek (Lincoln, NE) by using 125 microsatellite markers. The existence of a pair of twins was determined by comparing the individual genotypes of the F2 offspring within family. Mutations were detected by comparing the genotypes of the parents to that of the offspring. Mutation rate was calculated as the ratio of the number of mutations observed in the population to the total number of alleles in the population for 125 marker loci.

### Results and Discussion

After screening the 65 families, one pair of individuals with identical genotype information was found. The two female piglets had matching genotypes at 122 of 125 marker loci. Three markers were inconclusive due to missing genotype information for either of the twins. The probability of obtaining two progeny that were identical by chance for 19 marker loci (one per chromosome) from the parents of the twins was  $5.29 \times 10^{-23}$ . Therefore we believe this resulting twin pair is real. However we cannot rule out the possibility that genotyping errors caused this pair of individuals to be misidentified or that other pairs may not have been identified. Additionally, we cannot dismiss any error that may have caused the same DNA sample to be analyzed for both individuals.

The twinning rate seen here is much lower than a previous estimate (1) that reported four pairs of identical twins from 163 fetuses collected. The important distinction is that this previous estimate came from fetal examinations taken at day  $29 \pm 2$ . With the known competition between porcine embryos, estimates of twinning *in utero* may not reflect values at birth. Our observed twinning rate of one twin pair from 525 offspring is an original estimate of identical twin births in swine.

Three microsatellite mutations were identified after comparing the genotypes of the F1 and F2 individuals. A total of 134,565 alleles was examined. This gave an average mutation rate of  $2.23 \times 10^{-5}$  for microsatellites in the population. This is comparable to an earlier study (4) that reported a mutation rate of  $6 \times 10^{-5}$  for swine microsatellites.

### Acknowledgements

The QTL project and this work were supported by an industry consortium consisting of National Pork Producers Council, Iowa Pork Producers Association, Iowa Purebred Swine Council, Babcock Swine, Danbred USA, DEKALB Swine Breeders, PIC International Group, Seghersgenetics USA, and Shamrock Breeders. This work also was funded by the Iowa Agriculture and Home Economics Experiment

Station, Ames, project no. 3148, as well as by Hatch Act and State of Iowa funds. Comments and suggestions by Daniel Pomp, University of Nebraska, and Rohan Fernando, Iowa State University, are greatly appreciated.

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